

The Effect of Compaction and Biosolid Fertilizer on the Functional Morphologies in Young Plants

Research Thesis

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by

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Abstract

Restoration efforts in urban landscapes often include spreading seeds of native prairie species over compacted soils. Soil amendments such as fertilizers are frequently added to promote growth and success of the seedlings. Understanding the morphologies and success of young seedlings in stressful environmental conditions, like compacted soil, can indicate the success of the mature plant. However, little is known about young seedling traits, specifically root morphologies. We analyzed seedling traits of four species of prairie plants grown in artificially compacted soils. These species included two phylogenetically paired forbs, *Asclepias tuberosa* and *Ratibida pinnata*, and two phylogenetically paired grasses, *Schizachyrium scoparium* and *Andropogon gerardii*. In both cases, root morphologies differed between species in a pair. The seedlings were grown in one of three treatments or a control. The treatments consisted of soil amendments of half or quarter biosolid fertilizer, or a till treatment. Functional morphologies of the young plants were analyzed to determine differences between species or treatments as well as the interactions among treatments and species. *Andropogon gerardii* was expected to be the most successful species due to its spreading root structure overcoming the compacted soil. Additionally, the treatment with the greatest amount of biosolid was expected to grow the largest individuals, both above and belowground due to the additional nutrients. Averaging across all treatments, one species was not the most successful over the other species. The treatment with the greatest nutrient addition, the half biosolid, was not found to be the most successful. This result indicates nutrient overload and salt additions should be taken into considerations when adding soil amendments in restoration efforts. Future analyses need to be completed to understand the success of seedlings in stressful environments and how their traits can be indicative of longer-term success.

Introduction

Ecological research can benefit restoration efforts by providing critical information on species and their environments to ensure the most successful outcomes. Research on species-specific traits from plants grown in similar environments to the restoration area can support successful colonization in that area. Specifically, understanding traits during the progression from seed to seedling is crucial to ensuring establishment. The initial life stages of a plant are often matched with high mortality rates. **Although this part of the lifecycle is critical, scientists know very little about how functional traits of seedlings influence plant establishment and survival (Larson and Funke 2016).** Additionally, root morphology is critical to successful establishment of mature plants and little is known about how it can impact early life stages of plants. Knowing morphological impacts on plants during seed germination, emergence, and establishment can better the chance of success and establishment of mature species.

Understanding morphological impacts on seedlings grown in urban soils is particularly important in restoration projects that target urban lots. Oftentimes, a variety of grasses and forbs are used to restore the biodiversity in urban vacant lots. Understanding the differing success of these species in urban soils can provide information on the success of the restoration project. Compacted soils and the removal of topsoil are common in urban landscape development. This can present obstacles in restoration projects because soil compaction limits a plant's access to oxygen, water, and space to grow as well as disturbed nutrient cycles (Kozlowski 1999).

Compaction from urban landscape development can present a hinderance to germination and successful establishment of seedlings in urban restoration projects. Compaction in soil is defined as an increased density, increased resistance to penetration, and decreased amount of

space for water and air (Skinner et al. 2009). Scientists know soil compaction impacts traits in adult plants, but little is known about how soil compaction affects seed germination, emergence, and establishment, despite the importance of the germination stage to overall plant development (Alameda and Villar 2012). In restoration efforts, land managers are likely to start plants from seeds because it is an easy and cheap way to grow plants. However, there was found to be a variation in effect of soil compaction between species, soil type and compaction level (Skinner et al. 2009). By analyzing several functional traits of seedlings grown in compacted soil, we can understand how soil compaction affects the early life growth of native, prairie plants. These results can be helpful in restoration by making the effort as successful as possible.

Beyond these challenges due to compaction, removing topsoil in urban land development can also limit nutrients and remove organic matter from the soil. Oftentimes soil amendments are added to urban soils to combat this limitation. However, finding the balance in how much of the amendment can be difficult. By introducing soil amendments there must be enough nutrients added while not overloading the soil with too many nutrients or salt. High concentration soil amendments can sometimes lead to nutrient loss from runoff and leaching while high salinity can also impact germination and growth (Heyman et al. 2019). In the City of Columbus, Com-Til® (City of Columbus 2020), an organic compost made from biosolids of wastewater treatment plants, is applied to compacted, vacant lots of soil to improve seed establishment (Basta et. al 2016). . This product provides nutrients that plants require and is used as a fertilizer to enhance vegetation establishment on vacant lots. The effects of Com-Til® on seed development have not been explicitly measured previously (Basta et. al 2016, L. Weston *Unpublished*).

By understanding traits and establishment success of different species of seedlings, land managers can choose the species with the greatest germination and emergence success for their

restoration projects. Two phylogenetically paired forbs, *Asclepias tuberosa* and *Ratibida pinnata* will be analyzed as well as a two of phylogenetically paired grasses, *Schizachyrium scoparium* and *Andropogon gerardii*. Each species offers a different rooting habit to compare the differences in impacts of compaction. Among the four species we are testing, we expected that the spreading grass species, *Andropogon gerardii*, would be the most successful because its spreading root system will better overcome the compaction. Understanding the impacts of soil environments in restoration efforts can also contribute to a successful project. Here, we evaluate the effects of compacted soil and Com-Til® amended treatments on the functional morphology of commonly used prairie restoration species from the start of germination. We expected that plants treated with the highest amount of Com-Til® will show the largest growth in plant traits such as root biomass and shoot height.

Methods

STUDY SPECIES

We chose four species of native, North American prairie plants, each with differing root systems as our study subjects. This included *Asclepias tuberosa*, a tap rooted forb, *Ratibida pinnata*, a fine rooted, spreading forb, *Schizachyrium scoparium*, a bunch grass and *Andropogon gerardii*, a spreading grass. These were chosen based on previous work done with mature individuals of these species grown in compaction with similar treatments (L. Weston *unpublished*). These species represented two phylogenetically matched pairs with two forbs and two grasses. The pairing allows for comparison between genetically similar species. Despite the phylogenetic matching, the root morphology differed within each pair. We used the differing root morphologies to compare success within compact soil and the treatments. Although we had

four different species each with different root structures, root habits cannot be generalized from only these four species. Each species grew in artificially compacted soil with an added layer of one of three treatments or the no-amendment control.

SOIL COMPACTION TRIALS

Prior to designing a method of artificially compacting the soil, the soil mixture needed to be homogenized with half sifted subsoil and half sand. The subsoil was obtained from Waterman farm in Columbus, Ohio. Subsoil was sifted with a 2mm sieve to extract any large aggregates in the soil. The sifted soil was then fully mixed with sand using a cement mixer to make a 50/50 mixture. Next, we designed and tested a variety of procedures to artificially compact the soil. Our pots for this experiment were a 10.02 cm diameter PVC pipe cut into 14.5 cm pieces.

To obtain a soil resembling urban vacant lot environments, the soil needed to be artificially compacted using a uniform procedure. This procedure needed to compact the soil into the PVC pots to obtain a bulk density near 1.70 g/cm^3 . We chose this value because soils with high clay content and bulk densities of 1.7 g/cm^3 have been found to hinder plant growth (Moebius-Clune 2016). To stabilize the pot during compaction, a mold surrounding the pot was designed using QuickCrete. Soil was uniformly added before each round of weight drops. Circular “cookies” or wood cut-outs were made to slide into the pot on top of the soil to compact it on a level plane. These wood cookies would be taped together to an additional cookie nailed to a level piece of particle board for the weight to be dropped on. A weight would then be dropped from a height of 45 cm two times per soil layer onto the pot with the wood cookie board on top to compact the soil beneath it. The 6.1 kg weight consisted of a reinforced cardboard box with a weighted blanket inside. This process of adding soil, sliding in wood cookies, and dropping the

weight was completed until the compacted soil was near the top. This consisted of three 4 cm layers of soil and three rounds of weight drops per pot. The process as well as pictures of some of the materials are shown in Figure 1. This procedure obtained a bulk density of 1.70g/cm^3 with 12.1 cm of soil in depth.

SOIL AMENDMENT TREATMENTS

After developing a uniform compaction method, 80 pots were prepared with one of three soil amendment treatments or the control. The soil amendment treatments were a 2.5 cm layer on top of the compacted soil of either half biosolid, quarter biosolid, or till. These soil amendment treatments are meant to represent soil amendments used in restoration efforts. The half biosolid treatment consisted of a 2.5 cm layer on top of the compacted soil of a mixture of one-half Com-Til® and one-half subsoil. This soil amendment treatment consists of the highest nutrients while allowing for a buffer zone from the compact soil; it represents pre-planting soil amendment treatments where a mixture of Com-Til® and topsoil are spread across the site but not incorporated into the subsoil. The quarter biosolid treatment consisted of a 2.5 cm layer of one-fourth Com-Til®, one-fourth topsoil, and one-half subsoil. This soil amendment treatment was not as nutrient dense but still offers the buffer zone and represents the incorporation of a Com-Til®/topsoil mixture into the subsoil via shallow tilling. Initial germination was unsuccessful with a full Com-Til® treatment suggesting that salt concentrations were too high for a total Com-Til® treatment. The till treatment consisted of a 2.5 cm layer of only subsoil. This is representative of land managers tilling the compact soil before planting seeds for their

restoration project. Finally, the control consisted of 0.5 centimeters of subsoil. Adding a small amount of subsoil represents the fine layer of soil that has been disturbed on top of the compact urban soils and was meant to guard against complete germination failure in the control treatment.

GROWTH, GERMINATION, AND WATER SCHEDULE

After the soil amendment treatment or control layers were added, the pots were seeded. The seeds were ordered from Ohio Prairie Nursery in Hiram, Ohio in 2019. To reduce pathogen induced mortality, all seeds were sprayed with 5% bleach solution before seeding. The pots were seeded with one of our four species by placing ten seeds on top of the soil amendment treatment or control layer and then mixed into the soil. There were five replicates of all four species with each soil amendment treatment groups and the control leaving 80 pots total. All pots were watered with 100mL of water.

The next day, all 80 pots were put into a Conviron TCR30 growth chamber (Pembina, ND) for nine weeks. The growth chamber had a photoperiod of 12 hours with a 25°C day and a 15°C night. The pots were uniformly watered every other day with either 50mL or 100mL depending on the dryness of the soil. The number of germinated seeds per pot was counted for the first nineteen days in the growth chamber. After 18 days germinating in the growth chamber, the shoots were cut from all but one of the emerged seedlings to reduce the density to one plant per pot.

HARVEST AND MEASUREMENTS

After 63 days in the growth chamber, we measured the maximum height of each plant. A medium sized leaf from each pot was then removed and imaged with an Epson XYZ scanner and analyzed with WinFolia (Regents Instruments, Quebec, Canada) for leaf area. The rest of the above ground material for each pot was also cut with scissors. The leaf and above ground mass were weighed fresh and put into a 60C drying oven for at least 24 hours. The dry weight of the leaf and dry above ground mass was recorded the next day.

The below ground biomass was carefully washed with water to clean the soil from the roots. The cleaned roots were then put into water in plastic bags and put into the refrigerator until all samples had been washed (within 3 days). To prepare root biomass for root trait quantification, we took roots from the bags, gently patted them dry, and then left them to air-dry at room temperature for exactly three minutes. The roots were then weighed, stained with methylene blue to be imaged with an Epson XYZ scanner and analyzed with WinRhizo (Regents Instruments, Quebec, Canada) to estimate root diameter and length (Roumet et al. 2008). The roots were then placed in a 60C drying oven for over 72 hours and then weighed to quantify total belowground biomass.

STATISTICAL ANALYSIS

All results were analyzed with R version 1.4.1103 (RStudio PBC, 2021). Analyzed traits include dry aboveground biomass, dry belowground biomass, plant height, total root length, average root diameter, and seeds germinated. Root mass ratio was calculated as the ratio of belowground biomass to total biomass. Specific root length was calculated by dividing total root length by belowground biomass. Finally, specific leaf area was calculated by dividing leaf area

by leaf dry mass. *Ratibida pinnata* did not have enough species survive past germination to perform statistical analyses on any traits other than seeds germinated.

For germination analysis, we conducted a two-way ANOVA to test for differences in the number of seeds germinated based on species identity, on soil amendment treatment, and their potential interactions. For the grasses *Schizachyrium scoparium* and *Andropogon gerardii*, we conducted a two-way ANOVA to test for trait differences based on soil amendment treatment, species, and their potential interaction for all traits. For *Asclepias tuberosa*, we lacked a phylogenetically paired species comparison, so we investigated soil amendment treatment differences only, using one-way ANOVA. Tukey pairwise comparisons were used for all traits following a significant effect of soil amendment treatment, species, or the treatment x species interaction. In all cases we used a critical alpha value of 0.05.

Results

Analyses on the number of seeds germinated included data from all four of our study species. The interaction ANOVA model was a better fit to the data, and there was a significant effect of the interaction between soil amendment treatment and species (Table 1). For *Andropogon gerardii*, 50% more seeds germinated in the half biosolid treatment than in the control (Fig. 2). This result was not present in the other three species. Instead, there were no differences in the number of germinated seeds between the control treatment and the half biosolid treatment within each of the other three species. However, there were 34.5% more seeds germinated in *Andropogon gerardii* than *Schizachyrium scoparium* in the half biosolid treatment. This was the only significant difference between soil amendment treatments and between phylogenetically paired species. Among the species, *Schizachyrium scoparium* had significantly

fewer germinated seeds than the other three species. *Schizachyrium scoparium* had 47.2% fewer germinated seeds than *Andropogon gerardii*, 33% fewer than *Ratibida pinnata*, and 26.8% fewer than *Asclepias tuberosa*. The two forbs, *Asclepias tuberosa* and *Ratibida pinnata*, had similar numbers of seeds germinated, averaging around 64% and 52% of their seeds germinating, respectively.

For forbs, variation in above and below ground traits was analyzed for *Asclepias tuberosa* only because most *Ratibida pinnata* plants died within 14 days after germination. For *Asclepias tuberosa*, there were significant differences between soil amendment treatments in root diameter, root length, and height (Table 2). Root diameter differed between the control and the quarter biosolid treatment as well as between control and till (Fig. 3H). The quarter biosolid treatment led to 25.3% bigger root diameters than the control ($p=0.016$), and the till treatment yielded 30.0% bigger diameters than control ($p=0.014$). Root length also differed among soil amendment treatments (Fig. 3F). The half biosolid treatment yielded 56.9% longer roots than quarter biosolid ($p=0.011$) and 74.6% longer roots than till ($p=0.0063$). There was a slight difference between the control and quarter biosolid treatments, with plants in control conditions having 46.8% longer roots ($p=0.052$). Control plants also had 68.6% longer roots than the till treatment ($p=0.023$). Height differences among soil amendment treatments were only marginally significant ($p=0.057$; Table 2; Fig. 3B), but plants in the till treatment were 60.7% shorter than the control. There were no significant differences among soil amendment treatments for *Asclepias tuberosa* aboveground biomass, specific leaf area, belowground biomass, and specific root length (Table 2; Fig. 3).

For our dataset of grass seedling traits, the non-interaction ANOVA model was a better fit for analyses on all traits (i.e., there were no significant interactions between treatment and species). Significant differences were found among soil amendment treatments in above ground

biomass, and height (Table 3). There were also significant differences among species in dry above ground biomass and height as well as root diameter and root length. Aboveground biomass of grasses differed by soil amendment treatment and species (ANOVA $p=0.014$ and $p<0.001$ respectively). Averaged across soil amendment treatments, *Andropogon gerardii* had 60.7% more aboveground biomass than *Schizachyrium scoparium* ($p<0.001$; Table 3; Fig. 4A). Averaged across species, the quarter biosolid treatment had more aboveground biomass than control and till (Tukey $p=0.027$ and 0.027 , respectively), with no other differences among soil amendment treatments. Height differed by soil amendment treatment and species in the grass subset (ANOVA $p=0.039$ and $p<0.0001$ respectively). *Andropogon gerardii* was 38.2% taller than *Schizachyrium scoparium* (ANOVA $p<0.001$, Table 3; Fig. 4B). Between both grasses, quarter biosolid treatment was 35.2% taller than the control (Tukey $p=0.044$), with no other differences among soil amendment treatments. Across all soil amendment treatments, root diameter differed between the grass species with *Andropogon gerardii* having 14.1% thicker roots than *Schizachyrium scoparium* (ANOVA $p=0.017$; Table 3; Fig. 4F). Similarly, root length differed between the grass species with *Andropogon gerardii* having 61.9% longer roots than *Schizachyrium scoparium* (ANOVA $p<0.001$). There were no differences among soil amendment treatments in root length, specific leaf area, dry belowground biomass, or specific root length for grasses.

Discussion

The number of germinated seeds over 18 days varied by species and by soil amendment treatment as shown in Figure 2. *Andropogon gerardii* was the only species with variation in the number of germinated seeds by soil amendment treatment. This suggests that *Andropogon*

gerardii germination in urban, compact soils could be dependent on soil amendments. *Asclepias tuberosa* and *Ratibida pinnata* had similar and relatively high germination success. This could be evidence for forbs being more tolerant of compact soil conditions than grasses regardless of soil amendments during germination. *Schizachyrium scoparium* had low germination across all soil amendment treatments, and this response was insensitive to soil amendments. Other studies have found *Schizachyrium scoparium* to have similar emergence success regardless of the soil but intolerance to poorly aerated soils (Hitchmough et. al, 2004). Our results suggest that, in restoration efforts, *Schizachyrium scoparium* should be seeded in higher numbers to offset the low germination percentage.

In *Asclepias tuberosa*, there were significant differences in the height, root length, and root diameter based on soil amendment treatment. For each of these traits, the till treatment differed significantly from the control, with half and quarter biosolid treatments having intermediate results (Fig. 3-B and 3-H). Root length was the only exception to this with half biosolid having the longest roots of all soil amendment treatments (Fig. 3-F). The thick, tap root that *Asclepias tuberosa* normally produces may have offered an opportunity for a greater degree of variability in root morphology overall as a response to local conditions. Shorter and thicker roots in the till treatment may reflect seedlings optimizing the uncompacted soil layer instead of penetrating the compacted soil. While this may have avoided the compact soil environment, the plants grown in till also offered the shorter above ground height. Perhaps their height was reduced to avoid rooting further into the compact soil. Data on the effects of soil amendments on seedling traits is limited, so this causation is speculative. Additionally, *Asclepias tuberosa* seedlings grown in the control presented the second tallest aboveground height as well as the longest roots. However, these plants also presented the thinnest root diameters. Nutrient

availability may have caused this effect as the till treatment should offer more nutrients than the smaller layer with the control. Mature plants grown in soils with soil amendments often have greater root biomass (Thomsen 2006). The seedlings grown in the half biosolid treatment did have the longest roots but offered intermediate effects for other traits.

In the grass subset, aboveground biomass and height had significant differences in both soil amendment treatment and in species. For both *Andropogon gerardii* and *Schizachyrium scoparium*, the quarter biosolid treatment presented the plants with the greatest aboveground biomass over the other treatments. This differed significantly from the control and the till treatment (Fig. 4-A). This result may be indicative of the addition of nutrients and water holding capacity within the quarter biosolid treatment. More nutrients and water capacity would allow the plant to grow more mass above ground. However, the half biosolid treatment was intermediate among all other groups indicating a possible nutrient overload. Additionally, nutrient additions have not been found to impact aboveground biomass in prairie grasses (Tilman and Wedin 1991). Height also showed significant differences among soil amendment treatments in the grasses. As shown in Figure 4-B, the quarter biosolid treatment resulted in the tallest *Schizachyrium scoparium* plants and nearly the tallest *Andropogon gerardii* plants. Soil amendments and nutrient additions have been found to increase height in mature *Schizachyrium scoparium* plants, but this result was not consistent with mature *Andropogon gerardii* plants in previous studies (Tilman and Wedin 1991). Here, the quarter biosolid treatment was significantly taller than the control, similarly suggesting that added nutrients play a role in above ground growth for seedlings.

Root length and root diameter showed significant differences between the two grass species. *Andropogon gerardii* exhibited longer and thicker roots relating to its spreading root

structure while *Schizachyrium scoparium* exhibited shorter and thinner roots. This could also reflect differing responses between species to the compacted soil. While *Andropogon gerardii* exhibited larger belowground root traits, it is typical of *Schizachyrium scoparium* to produce substantial belowground biomass relative to its smaller aboveground biomass in mature plants (Weaver, 1991). However, this is not exhibited in our study with no significant difference in belowground biomass or in root mass ratio between the grass species. This could be indicative of a difference in traits between young and mature plants but literature on the belowground traits of young plants is limited.

Overall, we identified trait variability in these young plants depended on the species, root structure, and nutrient availability from soil amendments. There was not one species or soil amendment treatment that was consistently the most successful. The half biosolid treatment was not found to be the most successful and project managers should consider the potential negative effects of nutrient overload as well as salt concentrations when adding soil amendments in restoration efforts. Nutrient enrichment in the soil from biosolid amendments may also have community-wide negative impacts by encouraging aggressive, weedy species (Weston et. al Unpublished). While *Asclepias tuberosa* was the most successful in number of seeds germinated, above and below ground traits differed among soil amendment treatments. Future studies linking morphological traits of young, prairie plants to mature plant success are necessary for successful restoration efforts. Additionally, further investigations of seedling responses to stressful environmental conditions are needed to ensure successful establishment.

Figures and Tables

Table 1: Statistical analysis of the number of germinated seeds: ANOVA results from analysis of seed germination for all species and soil amendment treatments. Bold terms are those with p-values less than 0.05.

Source	Degrees of Freedom	Mean Squared	F-Value	Pr(>F)
Treatment	3	1.90	0.69	0.565
Species	3	80.90	29.15	<0.001
Treatment: Species	9	7.20	2.60	0.013
Residuals	64	2.78		

Table 2: Statistical analysis of *Asclepias tuberosa* traits: ANOVA results from analyses of all *Asclepias tuberosa* traits. Bold terms are those with p-values less than 0.05.

Trait	Source	Degrees of Freedom	Mean Squared	F-Value	Pr(>F)
Aboveground Biomass	Treatment	3	0.0001	1.35	0.312
	Residuals	10	0.00009		
Height	Treatment	3	13.56	3.53	0.057
	Residuals	10	3.85		
Specific Leaf Area	Treatment	3	5914	0.25	0.859
	Residuals	10	23521		
Belowground Biomass	Treatment	3	345.90	1.93	0.188
	Residuals	10	179.10		
Root Mass Ratio	Treatment	3	0.00000002	0.17	0.915
	Residuals	10	0.0000001		
Root Length	Treatment	3	3382	9.57	0.003
	Residuals	10	353		
Specific Root Length	Treatment	3	3.25	3.41	0.061
	Residuals	10	0.95		
Root Diameter	Treatment	3	0.016	7.18	0.007
	Residuals	10	0.002		

Table 3: Statistical analysis of grass species' traits: ANOVA results from analyses of *Andropogon gerardii* and *Schizachyrium scoparium* traits. Bold terms are those with p-values that are less than 0.05

Trait	Source	Degrees of Freedom	Mean Squared	F-Value	Pr(>F)
Aboveground Biomass	Treatment	3	0.0003	4.28	0.014
	Species	1	0.002	31.78	<0.001
	Residuals	27	0.00006		
Height	Treatment	3	123	3.21	0.039
	Species	1	776.6	20.25	<0.001
	Residuals	27	38.3		
Specific Leaf Area	Treatment	3	1258	0.28	0.837
	Species	1	1	0.00	0.986
	Residuals	27	4448		
Belowground Biomass	Treatment	3	523.8	2.14	0.118
	Species	1	335.9	1.38	0.251
	Residuals	27	244.3		
Root Mass Ratio	Treatment	3	0.0000002	0.18	0.909
	Species	1	0.000002	1.39	0.249
	Residuals	27	0.000001		
Root Length	Treatment	3	1746	1.93	0.148
	Species	1	23767	26.30	<0.001
	Residuals	27	904		
Specific Root Length	Treatment	3	7.24	0.69	0.568
	Species	1	19.66	1.86	0.183
	Residuals	27	10.55		
Root Diameter	Treatment	3	0.004	1.29	0.299
	Species	1	0.019	6.42	0.017
	Residuals	27	0.003		

Figure 1: Artificial compaction design: A – Picture of compaction design including adding the soil, the weighted box, the wood cookies, the number of drops, and the height. B – Picture of wood cookies on the level plane. C – Picture of the bucket filled with soil, the weighted blanket box, the stabilizing QuikCrete mold, and the level plane with wood cookies.

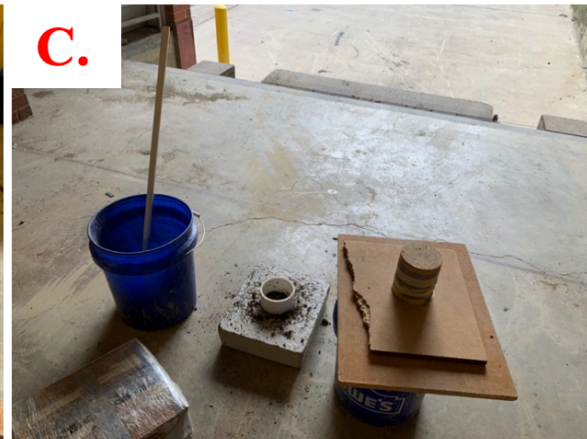
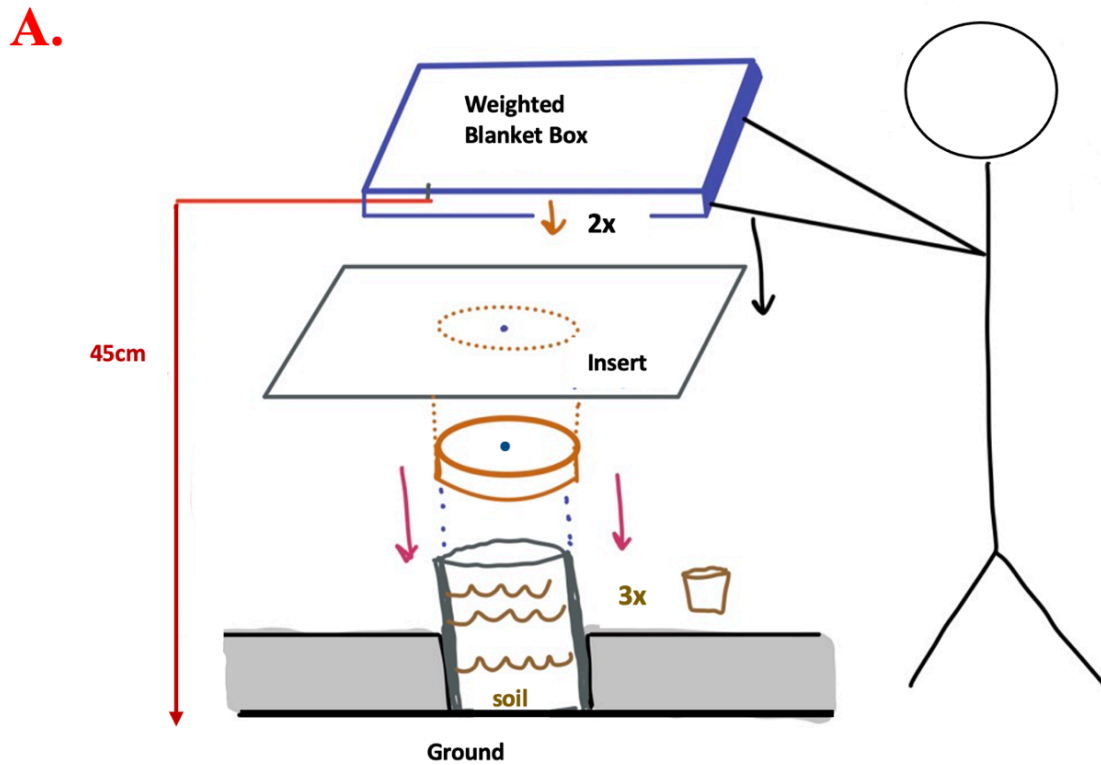


Figure 2: Number of germinated seeds: The number of germinated seeds in the first 18 days among all species and soil amendment treatments. The black line represents the median, the grey box represents the interquartile range, and the lines above and below the grey box represent the maximum and the minimum across replicates. The x-axis displays the soil amendment treatment and species respectively. “1/2” represents the half biosolid treatment, “1/4” represents the quarter biosolid treatment, “Cont” represents the control, and “Till” represents the till treatment. “ANGE” represents *Andropogon gerardii*, “ASTU” represents *Asclepias tuberosa*, “RAPI” represents *Ratibida pinnata*, and “SCSC” represents *Schizachyrium scoparium*.

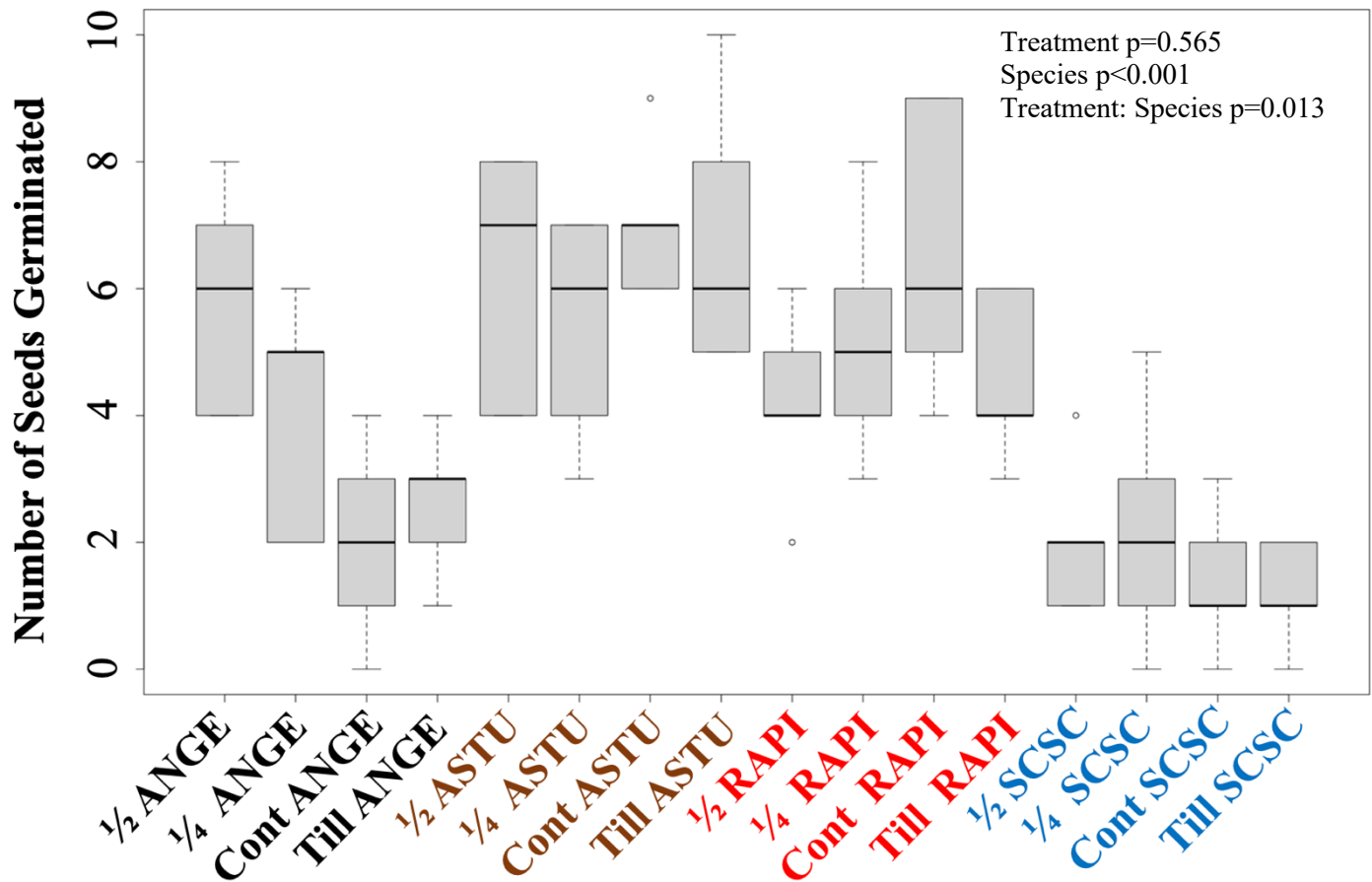
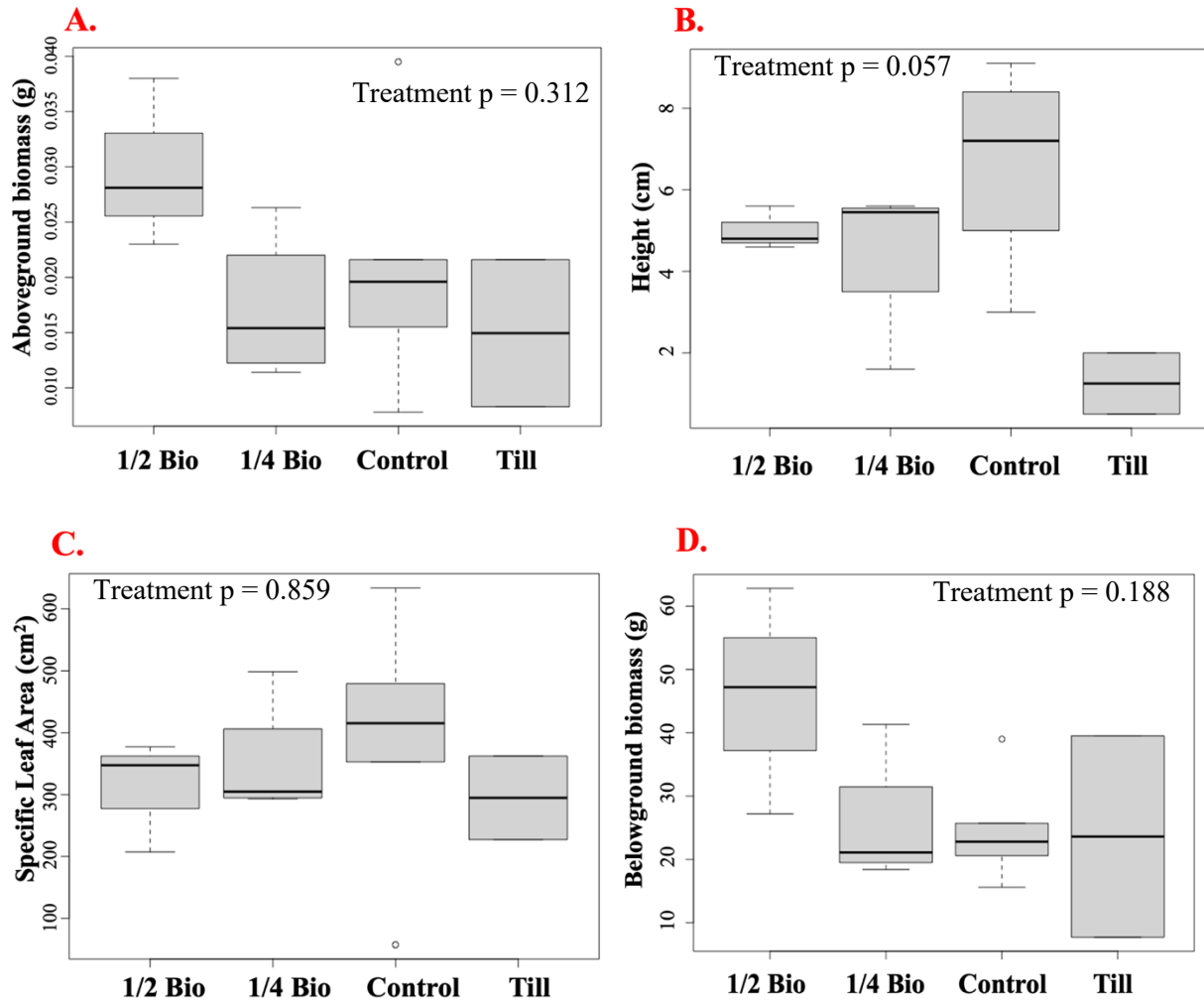
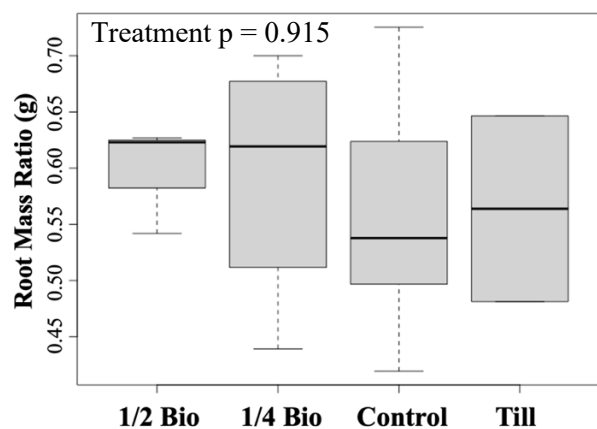


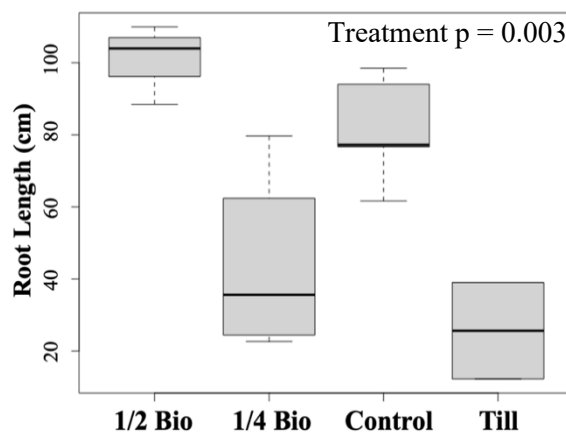
Figure 3: *Asclepias tuberosa* traits by treatment: For each soil amendment treatment, the dark line reflects the median, the gray boxes show the interquartile range, and the lines above and below the grey box represent the maximum and the minimum. Trait values shown are A) aboveground biomass, B) height, C) specific leaf area, D) belowground biomass, E) root mass ratio, F) root length, G) specific root length, and H) root diameter.



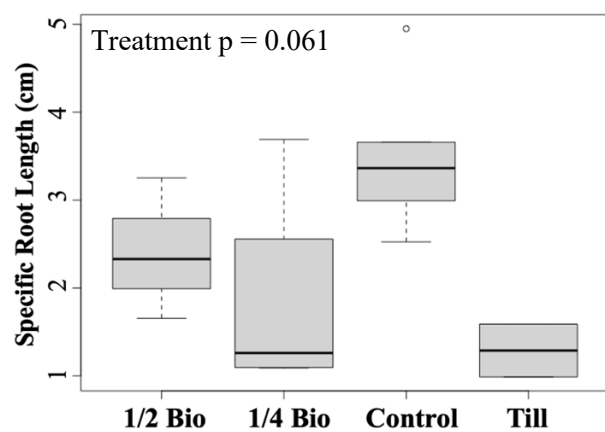
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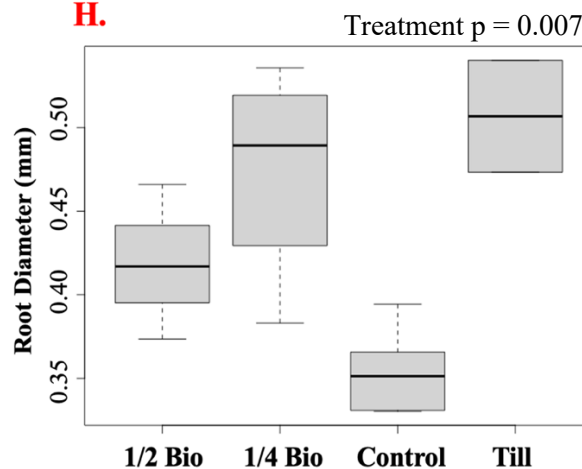
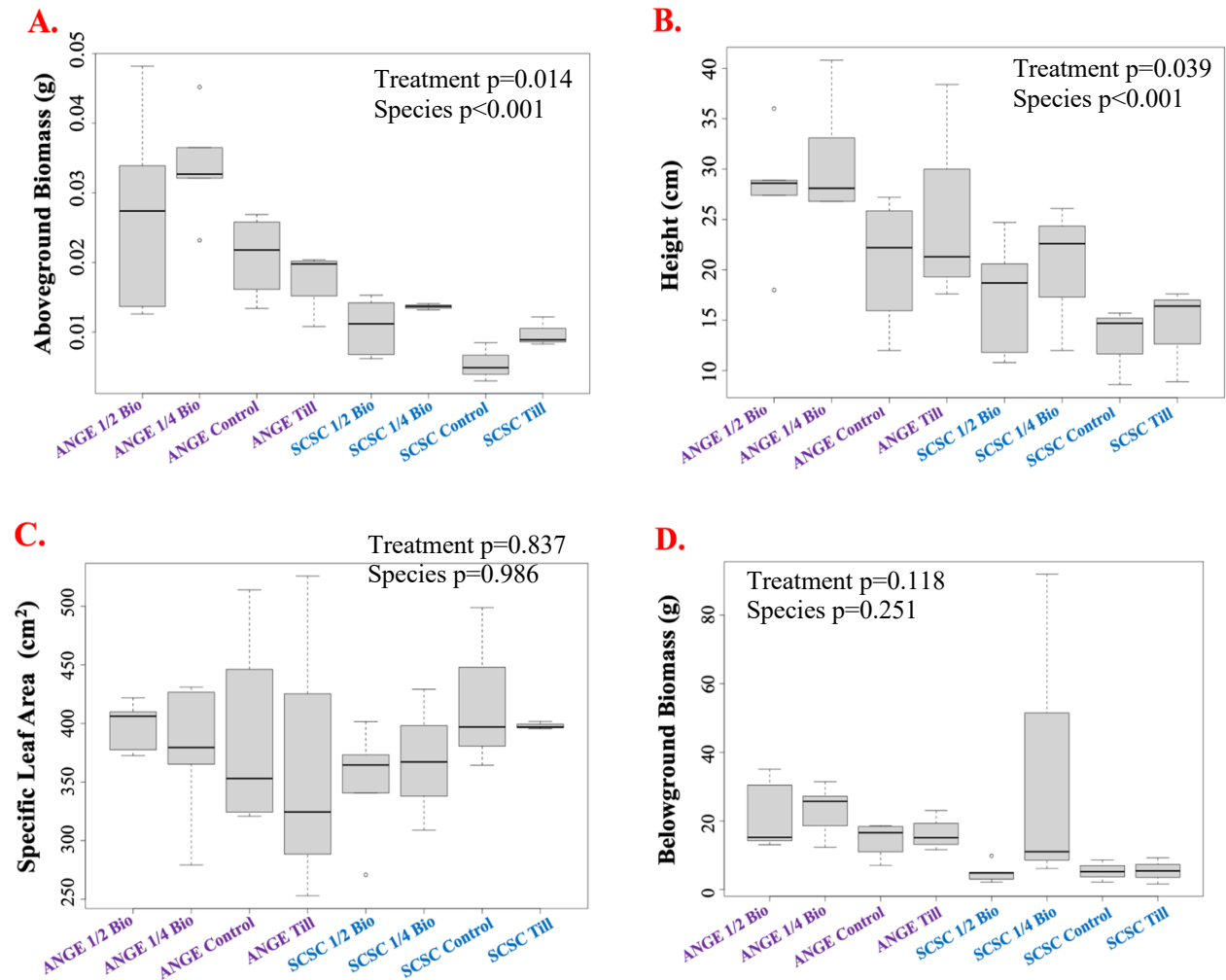
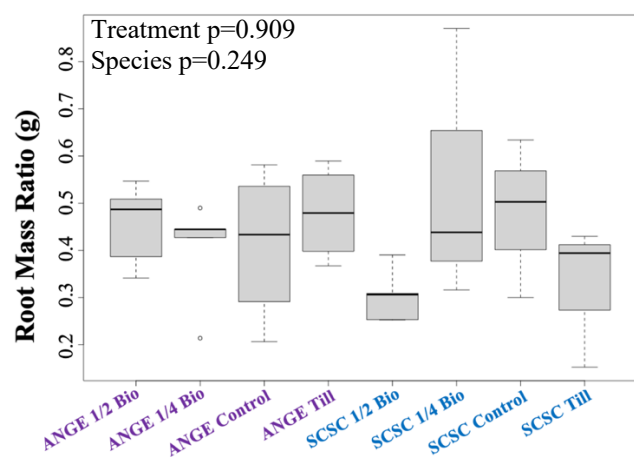


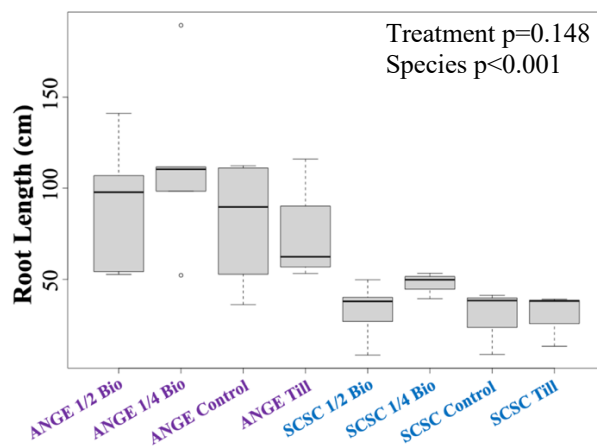
Figure 4: *Andropogon gerardii* and *Schizachyrium scoparium* traits by treatment: For each soil amendment treatment, the dark line reflects the median, the gray boxes show the interquartile range, and the lines above and below the grey box represent the maximum and the minimum. “ANGE” represents *Andropogon gerardii* and “SCSC” represents *Schizachyrium scoparium*. Trait values shown are A) aboveground biomass, B) height, C) specific leaf area, D) belowground biomass, E) root mass ratio, F) root length, G) specific root length, and H) root diameter.



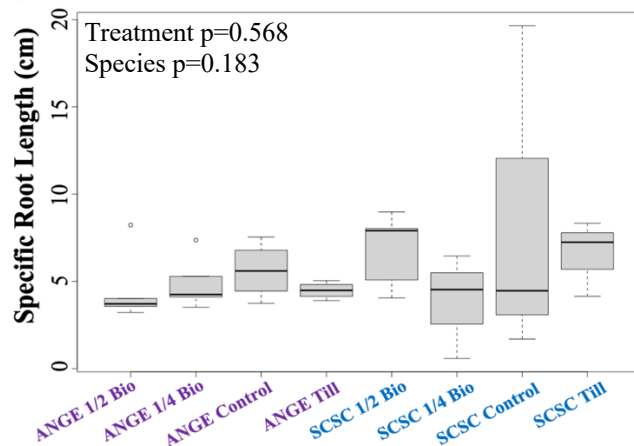
E.



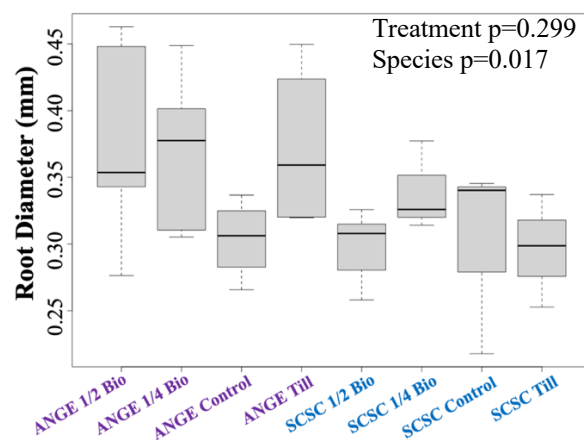
F.



G.



H.



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